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## Influence of different tillage, cropping sequence, mulching and fertility levels on microbial population

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**Abstract**

The field experiment was conducted at Agricultural Research Farm BHU Varanasi during *Kharif*, *Rabi* and summer season 2012-13 and 2013-14 to evaluate the microbial population under two tillage practices (reduced tillage and conventional tillage) four cropping sequence; (rice-wheat, rice-potato-green gram, maize-mustard-green gram and DSR-maize-green gram), two mulching treatments; (mulching during *rabi* and no mulch) and two fertility levels (75% RDF +25% N through FYM and 100% RDF) with three replications in Split Plot Design. The results revealed that reduced tillage significantly increased the population of Bacteria  $\times 10^5$  CFU  $g^{-1}$ , Actinomycetes  $\times 10^5$  CFU  $g^{-1}$  and Fungi  $\times 10^5$  CFU  $g^{-1}$  in soil over the conventional tillage. Significant increase of Bacteria  $\times 10^5$  CFU  $g^{-1}$ , Actinomycetes  $\times 10^5$  CFU  $g^{-1}$  and Fungi  $\times 10^5$  CFU  $g^{-1}$  in soil under maize-mustard-green gram cropping sequence were noticed over rest of the cropping sequences. Mulching significantly increased the population of Bacteria  $\times 10^5$  CFU  $g^{-1}$ , Actinomycetes  $\times 10^5$  CFU  $g^{-1}$  as well as Fungi  $\times 10^5$  CFU  $g^{-1}$  as compared to No mulch. Similarly, INM comparing 75% RDF + 25% N through FYM significantly improved the population of Bacteria  $\times 10^5$  CFU  $g^{-1}$ , Actinomycetes  $\times 10^5$  CFU  $g^{-1}$  as well as Fungi  $\times 10^5$  CFU  $g^{-1}$  over 100% RDF during both the years, all these practices improved soil fertility, soil health as well soil quality.

**Keywords:** Tillage, cropping sequence, mulching, fertility and microbial population

**Introduction**

Microorganisms in the soil robustly manipulate soil processes (Garbeva *et al.*, 2004) <sup>[5]</sup>, play key roles in the breakdown of organic matter, the cycling of carbon and nitrogen and the formation and maintenance of soil structure. Hence, the constituents of soil microorganisms, like as microbial and microbial population diversity, have frequently been recognized as perceptive indicators of biological index for maintaining soil health and superiority (Bending *et al.*, 2002) <sup>[1]</sup>. Unsustainable agricultural practices showed extreme soil erosion which can enhance physical and chemical degradation (Lal *et al.*, 1997) <sup>[2]</sup>. In response to the deterioration of regional soil quality in the Loess plateau, there has been a ongoing shift from conventional tillage towards reduced tillage practices such as zero tillage, crop residue incorporation and crop rotation. These production principles have resulted in optimistic effects on crop yield and soil physical and chemical properties also robustly influence the size, composition, multiplicity and function of soil microbial population resulting in considerably distorted soil processes. (Zero tillage has augmented soil organic carbon in the surface layer as well as build microbial biomass C (MB-C) in as little as five years and significantly enhanced soil microbial activity and diversity additionally). Conservation tillage system in which at least 30% of crop residues are left in the field and is an important conservation practice to decrease soil erosion. The benefit of conservation tillage practices over conventional tillage consist of reduction in cultivation cost (Parsch *et al.*, 2001) <sup>[9]</sup>; allowing crop residues to act as an insulator and reducing soil temperature instability; enrichment of soil organic matter; conserving soil moisture. Conventional tillage is not effective in improving microbial community as much compared to conservation tillage.

The rice-wheat system required high amount of nutrients to attain their possible yields (Hegde and Pandey, 1989) <sup>[9]</sup> but it is too expensive to poor and subsistence farmers of the state. Although, both crops in rice-wheat system are nutrient exhaustive, and result in reduced the soil-organic carbon and decline in soil health. Hence, this system has started showing symbols of low energy due to indiscriminate use of chemical fertilizer and excessive use of pesticides and irrigation water. The soil health and quality of water are deteriorating day by day which lead to the declining of crop productivity. Therefore, nutrient recycling in the soil-plant bionetwork is an indispensable component of sustainable productive agricultural enterprise. Declining factor productivity, Worldwide energy crises and price appreciation of chemical fertilizers has led to current importance on supplementation or replacement of chemical fertilizers with organics (Prasad, 2005) <sup>[6]</sup>.

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Organic manures and biofertilizers being less expensive, easily available and ecologically projected are known to improve soil fertility and crop yield. Plant nutrition by organics is a practice adopted by the farmers from ancient times. Organic sources of plant nutrients were efficient in improving wheat productivity, soil properties and biological activity (Singh *et al.* 2003) [11]. Organics also boost the efficiency of useful nutrients (Singh *et al.* 2012) [5]. Kumar *et al.* (2007) [7] found that the integration of FYM had useful effect on wheat.

Mulching is an useful method of changing crop growing environments in order to increase yield and develop product value by managing weed growth, decreasing soil temperature, conserving soil moisture, decreasing soil erosion, maintaining soil structure and improving organic matter content of the soil. Mulches are used for many reasons but water conservation and erosion control are the mainly major objectives for its use in agriculture in dry regions. Other reason for mulch use includes soil temperature alteration, soil conservation, nutrient addition, improvement in soil structure, weed management and crop quality improvement. Mulching reduces declining soil inabily by way of control runoff and soil loss, minimizes weed invasion and checks water evaporation. Thus, it facilitates more retention of soil moisture and helps in controlling temperature fluctuations, enhance physical, chemical as well as biological properties of soil, as it adds nutrients to the soil and finally enhances microbial population.

### Materials and Methods

The field experiment was conducted at Agricultural Research Farm, Department of Agronomy, Institute of Agricultural sciences, B.H.U., Varanasi Uttar Pradesh (India). The Agricultural Research Farm is situated in south-eastern part of Varanasi city on 25° 18' N longitude at an altitude of 128.93 m from the mean sea level. The experiment was laid out in the integrated farming System Research block of the Agricultural Research farm, which had homogeneous fertility and uniform textural make up. The experimental plot was typically a medium land which some time get submerged in rainy season and is suitable for rice in *Kharif* and also to the *Rabi* and summer crops. The field is well connected to the tube well irrigation system for frequent and timely irrigation. Varanasi (125° 18' N latitude and 83°03' E longitude) is situated in the north east plain zone in the eastern part of Uttar Pradesh, and it falls in the belt of semi arid to sub-humid climate. Varanasi lies almost in the middle of the Indo-gangetic alluvial plains; so, the soil is typically Gangetic alluvium (order-Inceptisols). In general, the alluvial soils are deep, flat, well drained and moderately fertile being low in organic carbon, available nitrogen, sulphur and zinc, and medium in available phosphorus and potassium. The soil of experimental field was sandy loam in texture having its natural drainage in river Ganges. Before the start of the experiment, composite soil samples were collected with the help of soil auger and core sampler. The normal period for the onset of monsoon in this region is third week of June and it withdraws by the end of September or sometime first week of October. A shower of rain is often experienced in January and February. The initial fertility status of soil before conducting experiment was studied; Composite soil samples from 0-15 cm depth were randomly collected from the experimental field. All the possible technical safety measures as approved for standard soil sampling technique were been followed. The soil samples hence obtained from 0-15 cm were subjected to various

chemical and biological analyses to evaluate the field sites for analysis of soil biochemical properties and microbial population counts after harvest of each cropping sequences under two tillage practices (reduced tillage and conventional tillage), four cropping sequence; (rice-wheat, rice-potato-green gram, maize-mustard-green gram and DSR-maize-green gram), two mulching treatments; (Rice straw mulch 6 t ha<sup>-1</sup> and No mulch) and two fertility levels (75% RDF +25% N through FYM and 100% RDF) in Split Plot Design. The treatments were replicated thrice.

Three different specific medium mentioned underneath were prepared as per the recommendation for isolation of different microbes such as bacteria, fungi, actinomycetes, for the quantitative and isolation of the microbes, media were prepared in liquid form which later solidifies.

One litre of the medium was prepared and distributed in the 5 nos. of 250 ml conical flask, which contained 200 ml in each flask. For each medium 5 replicates were made. After distribution, the flask were plugged and sterilized in the autoclave at 15 PSI for 15 minutes. Then the media containing flask were kept for cooling and utilization in further experimentation. Ingredients of the specific recommended media used for the experiment were as follows:

- Nutrient Agar for bacteria as described by Gordon *et al.*, (1973)
- Rose Bengal Agar for fungi as described by Martin, (1950)
- Actinomycetes Isolation medium for by Corke & Chase (1956).

### Quantitative estimation of soil microbes

To estimate the number of soil micro flora, counts were calculated on the basis of serial 10 fold dilution technique, using the pour plate methods and replicate of 10 gm soil samples, and an appropriate dilution as described by Johnson and Curl, (1972). Ten g of air dried soil was taken from each soil sample and were sieved properly to discard all the foreign particles and added to 100 ml of sterilized distilled water to make a dilution of 10-1, from this dilution 10 ml of the aliquot was transferred to 90 ml of sterilized distilled water to make dilution of 10-2. Likewise, the soil samples were serially diluted (six fold series). Aliquots of 1 ml from dilution 10-5 were spread on nutrient agar, ashby's medium, okons medium and pikovskaya's medium for total bacterial, Azotobacter, Azospirillum and phosphate solubilizers counts respectively, and from 10-4 on Rose Bengal Agar for fungi and Kenknights medium for actinomycetes. Each dilution was spread onto five replicate. The number of colonies forming on each medium was counted at 48-72 hrs for bacteria and after 120 hrs for fungi and actinomycetes, after incubation at 32 ± 2 °C for bacteria and 25 ± 2 °C for fungi and actinomycetes. Colony forming units per g of soil (CFU g<sup>-1</sup>) was calculated using the equation.

$$\text{CFU g}^{-1} = \frac{\text{No. of colonies}}{\text{Volume plated (ml)}} \times \text{Dilution factor}$$

From each dilution, colonies were picked at random and sub-cultured back onto selective media of isolation to obtain pure cultures. These pure isolates were stored after incubation and growth at 4°C for further analysis and identification.

### Result and Discussion

Critical examination of the data (Table 1) revealed lucid variation under two tillage practices; the reduced tillage

improving significantly Bacteria  $\times 10^5$  CFU  $g^{-1}$ , Actinomycetes  $\times 10^5$  CFU  $g^{-1}$  and Fungi  $\times 10^5$  CFU  $g^{-1}$  in soil over the conventional tillage. Microorganisms in soil strongly influence the organic matter decomposition and enhance soil fertility and microbial communities have been identified as sensitive indicator for biological diversity and improving soil health. The total bacteria, actinomycetes and fungi which were significantly higher under reduced tillage as compared to conventional tillage is attributed to minimum disturbance of soil and residue retention as advocated by Dongre *et al* (2017)<sup>[4]</sup>. The higher population Bacteria  $\times 10^5$  CFU  $g^{-1}$ , Actinomycetes  $\times 10^5$  CFU  $g^{-1}$  and Fungi  $\times 10^5$  CFU  $g^{-1}$  in soil were noticed under maize-mustard-green gram cropping sequence over rest of the cropping sequence but with respect to DSR-maize-green gram sequence, the differences could turn significant only during 2013-14 *i.e.* in the true sense the third year of experiment. This may be ascribed to the fact that maize based cropping sequence produced higher quantity of crop residue by which added more organic matter in soil to improve physical, chemical, biological properties of soil. Rice straw mulching @ 6 t  $ha^{-1}$  significantly increased Bacteria  $\times 10^5$  CFU  $g^{-1}$ , Actinomycetes  $\times 10^5$  CFU  $g^{-1}$  as well as Fungi

$\times 10^5$  CFU  $g^{-1}$  as compared to No mulch. Mulching with crop residue has been reported to enhance microbial population due to accumulation of nutrient into the soil that may result in enhanced microbial activity and soil fertility as well as productivity as reported by Bhagat (2016)<sup>[3]</sup>. Application of 75% RDF + 25% N through FYM significantly increased Bacteria  $\times 10^5$  CFU  $g^{-1}$ , Actinomycetes  $\times 10^5$  CFU  $g^{-1}$  as well as Fungi  $\times 10^5$  CFU  $g^{-1}$  population as compared to 100% RDF, The integrated nutrient management enhances organic matter in soil by which improve physical, chemical, biological properties and biological diversity, Singh *et al.* (2012)<sup>[12]</sup>. The application of organics in the form of FYM, Green manuring, brown manuring, Azolla can increases Bacteria Actinomycetes as well as Fungi counts in soil due to enhanced activity of urease and dehydrogenase in company with soil microbial biomass carbon. Consequently 25 % RDF can be substituted by FYM to improve the soil health and fertility correlated with significant increase in the microbial count *i.e.* Bacteria Actinomycetes as well as Fungi as compared to 100% RDF as quoted by Mallikarjun *et al.* (2018)<sup>[8]</sup>.

**Table 1:** Effect of varying tillage, mulching and nutrient management practices under different cropping sequences on population of Bacteria, Actinomycetes and Fungi after completion of each cropping sequence

Treatments	Bacteria $\times 10^5$ CFU $g^{-1}$		Actinomycetes $\times 10^5$ CFU $g^{-1}$		Fungi $\times 10^4$ CFU $g^{-1}$	
	2012-13	2013-14	2012-13	2013-14	2012-13	2013-14
<b>Tillage</b>						
Reduced	18.13	18.63	14.06	14.96	7.58	8.28
Conventional	16.87	17.57	13.52	13.84	7.16	7.66
S.Em $\pm$	0.25	0.33	0.14	0.33	0.14	0.17
CD (P = 0.05)	0.75	0.99	0.42	1.00	0.41	0.52
<b>Cropping Sequence</b>						
Rice-Wheat	16.24	17.56	13.37	13.75	6.85	7.54
Rice-Potato-Green gram	17.26	17.56	13.40	13.89	7.07	7.80
Maize-Mustard- Green gram	18.78	19.53	14.39	15.93	8.06	8.72
DSR-Maize-Green gram	17.73	17.75	14.00	14.02	7.52	7.82
S.Em $\pm$	0.35	0.46	0.20	0.47	0.19	0.24
CD (P = 0.05)	1.06	1.40	0.59	1.42	0.58	0.73
<b>Mulching</b>						
No Mulch	17.13	17.47	13.44	14.06	7.21	7.56
Mulch in rabi	17.87	18.73	14.14	14.74	7.53	8.38
S.Em $\pm$	0.26	0.18	0.20	0.22	0.11	0.14
CD (P = 0.05)	0.73	0.50	0.56	0.62	0.31	0.38
<b>Fertility</b>						
RDF	16.75	17.43	13.38	13.96	6.91	6.98
75% RDF + 25% N through FYM	18.25	18.77	14.19	14.83	7.84	8.96
S.Em $\pm$	0.26	0.18	0.20	0.22	0.11	0.14
CD (P = 0.05)	0.73	0.50	0.56	0.62	0.31	0.38

## Conclusion

The practice of reduced tillage significantly increased microbial population of Bacteria Actinomycetes as well as Fungi over conventional tillage during both the years. Among the four cropping sequences maize based sequences particularly the maize-mustard-green gram sequence markedly improved the population of Bacteria, Actinomycetes as well as Fungi over rest of the cropping sequences. Mulching with rice straw @ 6 t  $ha^{-1}$  during winter also significantly increased Bacteria, Actinomycetes as well as Fungi count in soil than No mulch. Similarly, the integrated nutrient management practices of 75% RDF + 25% N through FYM markedly enhanced Bacteria, Actinomycetes and Fungi population in soil over the 100% RDF during both the years. Here it may pointed out that under irrigated ecosystem of

Varanasi reduced tillage, mulching with rice straw mulch 6 t  $ha^{-1}$  and 75% RDF + 25% N through FYM in maize-mustard-green gram sequences significantly increased microbial population of Bacteria, Actinomycetes as well as Fungi population in soil.

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